

FIELD TESTS OF LARVICIDES AND WOUND PROTECTORS ON
NATURAL SCREWORM CASES, MENARD, TEXAS, 1938.
A SPECIAL REPORT

Treatments of natural screwworm cases in the field to test the larvicidal and wound protective properties of certain organic compounds were started in the vicinity of Menard, Texas, in the late spring of 1938. The materials to be tested were the organic compounds in dust form that showed promise as larvicides and wound protectors against the primary screwworm Cochliomyia americana C. & P. under laboratory and semi-field conditions. The object of the tests was to determine the practicability and effectiveness of promising chemicals under the natural variable factors they would be subjected to under range conditions, such as variations in severity and types of wounds, kind of animal, fly activity and weather conditions.

Tests were conducted on five selected ranches in the northwest part of Menard County. In these tests complete cooperation was obtained from owners and employees of the ranches for which members of the Menard station are grateful.

In the cooperative tests all materials, equipment, and labor needed in connection with the treatment of animals were furnished by the Bureau of Entomology and Plant Quarantine. The ranchers agreed to corral all "wormy" animals for each observation and to provide the necessary pens for their handling. The wounds of new infested animals were left untreated or were treated with benzol and the animals turned over to the Bureau of Entomology and Plant Quarantine for further treatment and observations. Such animals were confined in "worm-traps" and

were examined and treated when necessary until they healed.

In general, "wormy" animals were similarly cared for on the various ranches where tests were conducted. The animals were confined in open trap pastures where they could be found and corralled without much difficulty. Good grazing was available for animals on all but one of the ranches, but in this case additional food was provided in the form of hay and cotton seed cake. Water was provided for the animals in the trap pasture or in the pens. Watering places were inconveniently located in some instances. As a result, some of the weak or sick animals no doubt suffered from lack of water, which may have affected the rate of mortality.

Tests were started on May 31 and were discontinued on November 9. Animals were examined daily except Sundays and holidays until they healed. A rainy period prevented observations during the time July 20 to 25. However, because of exceedingly low fly activity due to almost continual rains, very few records were lost.

Five organic chemical dusts were used in the tests, namely, Diphenylamine, Menard Code Nos. 8, 9, 22 and 26. Pine Tar Oil, sp. gr. 1.065, or Bone Oil were used as standards of comparison.

The general procedure in treating the infested animals follows:

To keep individual records, each animal was given a numbered tag attached by means of a cord around the neck or horns. Notes were recorded on the kind, age, sex, length of hair and general condition of the animal. The position and dimensions of the wound, estimate of size, number, and species of larvae and the predisposing cause of the infestation were noted.

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In general, "kerry" animals were similarly cared for on the various ranches where tests were conducted. The animals were confined in open trap pastures where they could be found and corralled without much difficulty. Good grazing was available for animals on all but one of the ranches, but in this case additional food was provided in the form of hay and cotton seed cake. Water was provided for the animals in the trap pasture or in the pens. Watering places were inconveniently located in some instances. As a result, some of the weak or sick animals no doubt suffered from lack of water, which may have affected the rate of mortality.

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Wounds were treated with benzol using the technique recommended by the Bureau of Entomology & Plant Quarantine. Soiled wool or hair was closely clipped from the region of the wound with hand shears, and most of the larvae, particularly the larger ones, were removed with forceps before the material to be tested was applied.

Animals were generally taken at random for treatment with the various chemicals, although when several new untreated cases were available in the pens at the same time, some effort was made to make treatments with the different materials uniform as to type and severity of the wounds. Test materials were applied liberally to the wounds, and with the dusts the chemical was forced into wound pockets with the fingers or with the blunt end of a forceps. Some of the dust was also rubbed in the hair near the edge of the wound. Pine Tar Oil and Bone Oil were applied to the wound with paint brushes.

At each observation the wounds were carefully examined and the number of egg masses recorded. Notes on the condition of the wounds were made at about weekly intervals. After the original treatment, no further treatments were made unless the wounds became reinfested. In cases of reinfestation, after recording the age, number and species of larvae, the wound was treated with benzol, regardless of the size of larvae present, and liberally re-treated with the test material. Often newly-hatched larvae or larvae not definitely established were present in the wounds at the time of examination. Such wounds were not treated until the next examination, so as to allow sufficient time for toxic material in the wound to kill the larvae. In many such instances larvae

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failed to establish in dust-treated wounds.

A summary of the average healing time, average number of days protection against reinfestation, average number of re-treats for all animals that healed, and the percentage mortality of animals, is presented for the individual ranches in Tables Nos. 1 to 5. The same data combining the information for all ranches are condensed in Table No. 6.

From the standpoint of protection against reinfestation and average number of days to heal, it will be noted in Table No. 6 that the various dust compounds show superiority to Pine Tar Oil and Bone Oil, which were used as checks. It should be pointed out that tests with Menard Code Nos. 9, 22, 26, and Bone Oil were made during the last two months of the fly season and on relatively few cases. It is therefore difficult to compare results obtained on these materials with the results obtained on Diphenylamine, Menard Code No. 8 and Pine Tar Oil, which were tested more extensively.

Animals treated with Diphenylamine showed approximately the same rate of mortality as those treated with Pine Tar Oil, but with all other dusts the percentage mortality is lower than with Pine Tar Oil.

Special attention should be called to the average number of treatments required for animals treated with Pine Tar Oil as compared with those treated with dusts. The number of treatments with Pine Tar Oil was approximately twice as great as with Diphenylamine. Menard Code No. 8 showed slight superiority to Diphenylamine in average number of treatments required. Menard Code Nos. 22 and 26, although based

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on fewer cases, was about twice as effective as Menard Code No. 8. Of the dust compounds, Menard Code No. 9 proved least efficient from the standpoint of protection against reinfestations. However, as with Menard Code Nos. 22 and 26, the number of cases treated with this chemical is too limited to draw conclusions.

Bone Oil, by this method of analyzing the data, proved superior to Pine Tar Oil and equal to some of the dusts. However, the number of cases treated with Bone Oil is limited, and as shown by the average dimensions (Table No. 6) the wounds were smaller.

Table No. 7 presents the same data on protection against reinfestations as that contained in Table No. 6, with the addition of cases which required a re-treatment but on which records were not continued until the animal healed. These added cases include animals which died and those which were lost or otherwise disposed of before healing records were complete. As compared with Table No. 6, the results in general show fewer days protection against reinfestation and show a higher figure for the average number of reinfestations. The ratio of protection in the dust-treated animals as compared with that in animals treated with Pine Tar Oil or Bone Oil is, however, very nearly the same, showing a slightly greater advantage of the dust treatments over that of Pine Tar Oil and Bone Oil, as compared with results on healed animals as presented in Table No. 6.

Tables Nos. 8 to 13 show the frequency of infestations for all animals in which infestations were recorded, in relation to time intervals for the various materials used. The data are given for

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Tables Nos. 8 to 15 show the frequency of infections for all animals in which infections were recorded, in relation to time intervals for the various materials used. The data are given for

individual ranches in Tables Nos. 8 to 12, but may be studied best in Table No. 13, which combines the information from all of the ranches.

In summarizing these data, all reinfestations were considered whether from healed, dead, or from animals in which records for various reasons were incomplete.

In recording field notes, the age of larvae in each reinfestation was estimated. For example, larvae recently hatched but definitely established to those less than approximately 24 hours old were recorded as 1 day old. Larvae over 24 hours but not more than 2 days old were considered 2 days old, etc. In summarizing the field data, fairly accurate records could therefore be obtained showing the minimum amount of protection afforded by each treatment. In reality, however, the average age of larvae recorded as 1 day old was probably more nearly 1/2 days, and those recorded as 2 days old were more nearly 1-1/2 days old, etc., so that actually the percentage of infestations would be less than is indicated for the first days as shown in the tables. Considering the results from another angle, .5 percent reinfestations for Diphenylamine resulting in 0 days protection as recorded in Table No. 13 represents .5 percent reinfestations during the first 24-hour period after treatment, likewise 5.2 percent reinfestations giving 1 day protection represents reinfestations occurring on the second day after treatment.

By comparing the percentage of reinfestations in relation to periods of protection, the superiority of the dust treatments over Pine Tar Oil and Bone Oil is clearly shown. In Pine Tar Oil-treated

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Pine Tar Oil and Bone Oil is clearly shown. In Pine Tar Oil-treated

animals, 6.3 percent of the reinfestations resulted in 0 days protection, as compared with .5 percent for Diphenylamine and 0 reinfestations for Menard Code No. 8. The percentages of reinfestations in wounds treated with Menard Code Nos. 9, 22, and 26 indicate that these dust materials are about as effective as Diphenylamine or Menard Code No. 8.

Reinfestations resulting in a minimum of 1 day protection show a percentage of 24.5 for Pine Tar Oil as compared with a percentage of 5.2 for Diphenylamine and 2.9 percent for Menard Code No. 8.

By analyzing the data presented in Table No. 13 from a more practical standpoint, the following percentages of infestations may be expected when treatments of the various materials are made at 1, 2, 3, and 4 day intervals:

	<u>1 Day</u>	<u>2 Day</u>	<u>3 Day</u>	<u>4 Day</u>
Pine Tar Oil	6.3	30.8	50.3	61.2
Bone Oil	5.0	23.3	45.0	51.7
Diphenylamine	0.5	5.7	16.4	27.1
Number 8	0	2.9	11.6	24.0
Number 9	0	3.9	9.8	21.6
Number 22	0	4.2	12.5	22.2
Number 26	0.8	5.4	14.7	18.6

By noting the expected percentage infestations at various intervals of application, as based on results obtained in these tests, the dusts are several times as effective as Pine Tar Oil; Bone Oil shows slight superiority over Pine Tar Oil.

In connection with recommendations for frequency of treatment, it should be pointed out that observations indicate that larvae during

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Reinfestations resulting in a minimum of 1 day protection show a percentage of 24.5 for Pine Tar Oil as compared with a percentage of 5.2 for Diphenylamine and 2.9 percent for Menard Code No. 8. By analyzing the data presented in Table No. 15 from a more practical standpoint, the following percentages of infestations may be expected when treatments of the various materials are made at 1, 2, 3, and 4 day intervals:

	1 Day	2 Day	3 Day	4 Day
Pine Tar Oil	6.3	30.8	50.3	61.3
Bone Oil	2.0	32.3	45.0	51.7
Diphenylamine	0.5	2.7	16.4	27.1
Number 8	0	2.9	11.6	24.0
Number 9	0	3.9	9.8	21.8
Number 22	0	4.2	12.2	22.2
Number 26	0.8	2.4	14.7	18.6

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the first day of the infestation do very little damage to a wound. Likewise, larvae less than two days old, unless present in large numbers, apparently do little serious damage to the wound. The larger larvae 3 days old or older are of most concern. In considering this point, the percentage of infestations at various day intervals indicates that by treating every third day with Diphenylamine, only about .5 percent of the wounds would be infested with the larger, more destructive larvae and by treating every fourth day, 5.7 percent of the wounds would be infested with larvae 3 to 4 days old.

The larvicidal dusts used in these tests have proven, in preliminary tests, to be practically 100 percent effective as larvicides on larvae up to 3 days old. The percentage kill on older larvae is variable, but in general they are ineffective on larvae near maturity.

In field tests the value of the dust materials by increasing the kill of larvae is pronounced. As previously mentioned, all infested wounds were first treated with benzol, followed by treatment with the various test materials. Wounds treated with Pine Tar Oil, which has no larvicidal value, frequently had to be re-treated because of failure to kill 100 percent of the larvae, whereas 100 percent kill of larvae was obtained in almost all cases when the wounds were treated with benzol followed by treatment with dust materials. The advantage of increased larvicidal efficiency is of great practical importance from the standpoint of the average rancher, who often experiences difficulty in killing all larvae in wounds treated with benzol as well as other larvicides. Wounds in which large larvae are feeding, although the

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number of larvae present may be small, are readily subject to re-infestations. From the standpoint of efficiency in protection against re-infestations, it is therefore important that all larvae in wounds be killed.

Table No. 14 shows a summary of the performance of the various materials in relation to the different kinds of animals. In these tables all wounds were considered that became infested on animals that healed, died, or on animals for which records were incomplete. In some instances the number of cases treated with the various materials is very limited. However, in general the animals treated with Pine Tar Oil were more often infested than those treated with the dust compounds. There are no indications that the effectiveness of the dust materials over Pine Tar Oil is specific for any of the different kinds of animals on which these tests were conducted.

Although the type and severity of the wounds is probably a more controlling factor, the results show sheep to be more susceptible to reinfestations than any of the other animals, whereas hogs show the lowest incidence of reinfestation.

The relative number and percentage of cases of the various types of wounds for all animals treated is shown in Table No. 15. Cases in the eye (due largely to needle grass) and cases in the head (caused chiefly by fighting) were most frequent during the season.

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The effectiveness of the materials used on various types of wounds is shown in Table No. 16. All wounds were considered that became infested whether on healed, dead, or incompleated cases. Since the number of cases is so limited, very little conclusive information is indicated. In general, the dust compounds again show superiority over Pine Tar Oil. It is apparent, however, that the type of wound is important as a determining factor for the number of treatments required. Cases in the ear, for example, seem very susceptible to reinfestations even when treated with dust compounds, whereas the incidence of infestations in birth (navel) cases is low even when treated with Pine Tar Oil.

In developing a larvicidal dust for screwworm control, one object is to find a non-repellent material that will not prevent oviposition but will prevent infestations by killing the young larvae before they become established in the wound. Flies repelled from wounds already under treatment may be driven to other domestic animals or to wild animals, thereby starting new cases and building up the fly population in nature. A record was therefore kept of the number of egg masses collected on wounds treated with the various materials.

Frequently egg masses are lost between observations, for infestations often occur in wounds for which no egg masses have been recorded and on which no hatched eggs are in evidence at the time of the infestations. In instances of this kind, the number of egg masses deposited on the wounds was estimated on the basis of number of larvae

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present. Wounds treated with Pine Tar Oil in particular appear to lose egg masses readily, although this is also a frequent occurrence with dust-treated wounds.

Results showing the total and average number of egg masses deposited on the wounds of all animals treated with the various materials are presented in Table No. 17. Contrary to expected results, as based on preliminary field experiments, fewer egg masses were recorded on the wounds of dust-treated animals than on those treated with Pine Tar Oil. The average number of egg masses per wound was lower for Menard Code No. 8 than for wounds treated with Biphenylamine.

It may be worth while to theorize on the cause of the higher incidence of egg masses obtained on Pine Tar Oil-treated wounds. Several factors may be considered as an explanation for the results: (1) the dust materials may be repellent to screwworm flies; (2) the dust compounds used, because of their drying effect and more or less mechanical protective coating, may tend to decrease, neutralize or mask odors emanating from the wounds; (3) the fact that wounds treated with Pine Tar Oil were more often and frequently more severely reinfested no doubt was an important factor in increasing the incidence of egg masses on these wounds. It is well established that infested wounds are much more attractive to screwworm flies than are similar uninfested wounds.

It had been noted in earlier tests that the number of larvae present in reinfestations occurring in wounds treated with dust materials was smaller than in those treated with Pine Tar Oil. In order to gain some information on this point, an estimate of the number of larvae present in each reinfestation was recorded. The results are presented

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some information on this point, an estimate of the number of larvae present in each reinestation was recorded. The results are presented

in Table No. 18. Of the three materials, Pine Tar Oil, Diphenylamine, and Menard Code No. 8, the average number of larvae for each reinfestation was higher in Pine Tar Oil.

Since the number of cases treated with Bone Oil and Menard Code Nos. 9, 22, and 26 was limited and since, in some cases, treatments with these chemicals were not made during the same period as treatments with Pine Tar Oil, Diphenylamine and Menard Code No. 8, no conclusions can be drawn for these chemicals.

The number and percentage of screwworm cases involving the primary screwworm and secondary blowflies are presented in Table No. 19. As may be noted, the primary screwworm was involved in 87 percent of the infestations, whereas secondary blowflies were present in 13 percent.

The species and incidence of the various secondary blowflies is given in Table No. 20. It is interesting to note that the incidence of Cochliomyia macellaria Fabr. was highest in the original infestations, but Sarcophaga spp. were more frequently implicated in reinfestation involving secondary blowflies. It might be added that from all observations Sarcophaga larvae do little or no damage to a wound. C. americana larvae have difficulty in establishing in a pussy, suppurating wound. Sarcophaga larvae may clean out such a wound and render it suitable for the development of primary screwworms. Laboratory tests have shown that Sarcophaga larvae are very resistant to the dusts used as wound protectors. It is possible that Sarcophaga larvae may remove toxic dusts from the wounds and thus pave the way for C. americana infestations.

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infections.

The amount of chemical used for each treatment in the field no doubt will vary greatly with the individual making the treatment. The average in grams of the chemical dusts required per treatment in these tests was as follows:

Diphenylamine-----13.41

Menard Code No. 8--10.25

Menard Code No. 9-- 8.89

Menard Code No. 22-10.00

Menard Code No. 26- 7.91

Since Menard Code Nos. 9, 22, and 26 were used on a relatively small number of wounds and during only a part of the season, it should not be concluded that smaller amounts of these materials are required.

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Mercuric Code No. 9-8.89
Mercuric Code No. 22-10.00
Mercuric Code No. 26-7.91

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The chemical dusts used in the various experiments

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Mercuric Code No. 26-7.91 was highest in the various experiments.

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Mercuric Code No. 11.41 was highest in the various experiments.

Mercuric Code No. 22-10.00 was highest in the various experiments.

Mercuric Code No. 26-7.91 was highest in the various experiments.

Mercuric Code No. 9-8.89 was highest in the various experiments.

Mercuric Code No. 8-10.25 was highest in the various experiments.

Mercuric Code No. 11.41 was highest in the various experiments.

SUMMARY AND CONCLUSIONS

1. Field tests were conducted in the vicinity of Menard, Texas, during the screwworm season of 1938 to test the larvicidal and wound protective properties of certain organic compounds in dust form. The compounds tested were Diphenylamine and Menard Code Nos. 8, 9, 22, and 26. Pine Tar Oil and Bone Oil were used as standards of comparison as wound protectors.
2. Menard Code Nos. 9, 22, and 26 were tested during the last two months of the fly season and the number of cases treated is limited. Therefore, no conclusions can be drawn safely from the results. However, these dust compounds appeared to be equally or more effective as wound protectors than did Diphenylamine or Menard Code No. 8, and are worthy of further tests in the field.
3. On the basis of the average number of days protection for the initial treatment and the average number of days protection for all subsequent treatments, Diphenylamine proved superior to Pine Tar Oil. Menard Code No. 8 in these respects proved slightly superior to Diphenylamine.
4. On the basis of number of treatments required, Diphenylamine proved twice as effective as Pine Tar Oil. Menard Code No. 8 again proved slightly superior to Diphenylamine.
5. The percentage of infestations at various day intervals after treatment show Diphenylamine to be several times more effective in preventing reinfestations during the first 4 days after treatment than is Pine Tar Oil. On the same basis Menard Code No. 8 shows

SUMMARY AND CONCLUSIONS

1. Field tests were conducted in the vicinity of Newark,

Delaware, during the growing season of 1938 to test the following

and wound protective properties of certain organic compounds in

dust form. The compounds tested were Diphenylamine and Menard Code

Nos. 8, 9, 22, and 26. Pine Tar Oil and Bone Oil were used as

standards of comparison as wound protectors.

2. Menard Code Nos. 9, 22, and 26 were tested during the last

two months of the fly season and the number of cases treated in

listed. Therefore, no conclusions can be drawn solely from the

results. However, these dust compounds appeared to be equally or

more effective as wound protectors than did Diphenylamine or Menard

Code No. 8, and are worthy of further tests in the field.

3. On the basis of the average number of days protection for

the initial treatment and the average number of days protection for

all subsequent treatments, Diphenylamine proved superior to Pine Tar

Oil. Menard Code No. 8 in these respects proved slightly superior to

Diphenylamine.

4. On the basis of number of treatments required, Diphenyl-

amine proved twice as effective as Pine Tar Oil. Menard Code No. 8

again proved slightly superior to Diphenylamine.

5. The percentage of infestations at various day intervals

after treatment show Diphenylamine to be several times more effective

in preventing reinfestations during the first 4 days after treatment

than is Pine Tar Oil. On the same basis Menard Code No. 8 shows

distinct superiority over Diphenylamine.

6. The average healing time for wounds treated with Diphenylamine and Menard Code No. 8 was less than wounds treated with Pine Tar Oil. Of the two dust compounds, the wounds treated with Menard Code No. 8 healed faster than those treated with Diphenylamine.

7. Of the three materials, the percentage mortality of animals treated with Menard Code No. 8 was lowest. The percentage mortality of animals treated with Diphenylamine and Pine Tar Oil was about the same.

8. The larvicidal qualities of Diphenylamine and Menard Code No. 8 are especially valuable in wound treatment because supplementary action by the larvicidal dusts tends to compensate for incomplete kills with benzol, and larvae will seldom survive the double treatment. Whereas if benzol is followed by a non-larvicidal wound protector such as Pine Tar Oil, it often happens that some of the larvae survive.

9. On the basis of field tests, there are no indications that the effectiveness of dust materials is specific for certain kinds of animals.

10. The type of wound appears to be an important factor in determining the effectiveness of a wound protector.

11. The average number of larvae in reinfestations occurring in wounds treated with Pine Tar Oil was greater than that for Diphenylamine or Menard Code No. 8.

12. The average number of egg masses per wound on animals treated with Diphenylamine and Menard Code No. 8, contrary to expectations, was lower than on wounds treated with Pine Tar Oil. Of the two

distinct superiority over Diphenylamine.

6. The average healing time for wounds treated with Diphenyl-

amine and Menthyl Code No. 8 was less than wounds treated with Pine

Tar Oil. Of the two dust compounds, the wounds treated with Menthyl

Code No. 8 healed faster than those treated with Diphenylamine.

7. Of the three materials, the percentage mortality of animals

treated with Menthyl Code No. 8 was lowest. The percentage mortality

of animals treated with Diphenylamine and Pine Tar Oil was about the

same.

8. The larvicidal qualities of Diphenylamine and Menthyl Code

No. 8 are especially valuable in wound treatment because supplementary

action by the larvicidal agents tends to compensate for incomplete kills

with benzol, and larvae will seldom survive the double treatment.

Whereas if benzol is followed by a non-larvicidal wound protector

such as Pine Tar Oil, it often happens that some of the larvae survive.

9. On the basis of field tests, there are no indications that

the effectiveness of dust materials is specific for certain kinds of

animals.

10. The type of wound appears to be an important factor in deter-

mining the effectiveness of a wound protector.

11. The average number of larvae in reinfestations occurring in

wounds treated with Pine Tar Oil was greater than that for Diphenyl-

amine or Menthyl Code No. 8.

12. The average number of egg masses per wound on animals

treated with Diphenylamine and Menthyl Code No. 8, contrary to expecta-

tions, was lower than on wounds treated with Pine Tar Oil. Of the two

dust materials, the average number of egg masses per wound was lowest for Menard Code No. 8.

13. In addition to the primary screwworm, Cochliomyia macellaria Fabr., Sarcophaga spp., and Phormia regina Meig. were concerned in wound myiasis. The secondary blowflies were present in 13 percent of the cases treated. Of the secondary species, C. macellaria was most frequently involved in original infestations, whereas Sarcophaga spp. were implicated in a higher percentage of the reinfestations.

Respectfully submitted,

Edward F. Knipling
Assistant Entomologist.

January 18, 1939.

highest percentage of egg masses per wound was lowest
for Hensard Code No. 8.

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